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## Trapping Female Medflies (Ceratitis capitata) by Broadcast of Male Calling Song

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#### INTRODUCTION

Attractants for female medflies (1) are of particular interest in monitoring programs where sterile males are released to eradicate incipient populations (2). A female-targeted trap also could assist the monitoring of mass-trapping suppression efforts in areas of established populations (e.g., 3).

Mating in this species occurs primarily in leks where groups of males aggregate on the undersides of leaves near the tops of host trees (4). Each male occupies the underside of a leaf and emits a sex pheromone while vibrating his wings to produce a "calling song" at ca. 350 Hz (5). When a female lands on top of a leaf and climbs below, the male begins a series of visual displays, and if the female remains interested, he attempts copulation (e.g., 6).

To consider the feasibility of developing a female-targeted acoustic trap for medflies, we first observed that females landed and remained significantly longer at sites near speakers broadcasting a high-intensity calling song than at sites without song. These results led us to conduct several additional bioassays to determine whether broadcast calling song increased the percentages of females captured in adhesive traps.

#### MATERIALS AND METHODS

Insects. Ceratitia capitata pupae (standard medfly strain, sterilized under 14.5 kR radiation) were placed in screened cages on a 12-h light:dark light cycle and segregated by sex 2 days after emergence. Thereafter, the sexes were kept in separate rooms under temperatures of 24-26°C. Virgin females were used in bioassays 4-8 days after emergence.

Male medfly acoustic recording and playback. Recordings of 3-day-old male medflies during the first 4 h of photophase were made in an anechoic chamber with a Brüel and Kjær (B&K model 4145) microphone connected to a B&K measuring amplifier (model 2610), and the output was stored on a digital audio tape recorder (TEAC model DA-P1) and analyzed using the DAVIS insect sound analysis program (7).

Resting-time bioassay. The behavior of virgin female medflies was videotaped during morning and early afternoon periods of peak sexual activity. A 5-cm-diameter sheet of filter paper was placed vertically in front of the outlet of a funnel (20-cm diameter, with 2-cm diameter outlet) attached to a 25-cm-diameter, low-frequency speaker. Bioassays with different groups of 20-50 females were conducted for 2-h periods, alternating in 10-min observation periods with and without calling song at different intensities between 93 and 107 dB (Fig. 1).

Adhesive trap bioassays. In larger scale experiments, sound sources or silent mimics were placed at ca. 45-cm height, 30 cm from the ends of four  $61 \times 61 \times 152$ -cm Plexiglas chambers screened on the front and back. The sound sources faced the main part of the chamber (see Ref. 8 for description of the sound level distributions in the chambers). Medflies were captured in traps constructed from  $7 \times 7$ -cm pieces of (double-sided) yellow adhesive paper. Tests with 20-60 virgin females were run for 5-7 h beginning 2 h after the start of light phase. The percent capture in each trap was calculated by dividing the final count of females in that trap by the sum of all females trapped and all females remaining in the chamber at the end of the test.

#### **RESULTS**

Acoustic characteristics of male medfly calling song. The magnitudes and variations of the fundamental frequencies in the 11 recordings of calling songs from 3-day-old male medflies were within the ranges reported previously (9) for wild and sterile males. A representative sample can be found at cmave.usda.ufl.edu/~rmankin/medfly11.wav. The fundamental frequency is at 355 Hz, with lower intensity harmonics

Resting-time bioassay. No phonotaxis was observed when the 25-s sample of male calling song was broadcast in a continuous loop for 10-min periods at intensities between 93 and 107 dB sound pressure level (SPL). However, the resting time, <1.2 s/min in 10-min observation periods without calling song (55 dB SPL), increased to 2-10 s/min in observation periods when the SPL exceeded 105 dB (Fig. 2). A linear regression of the logarithm of resting

time on SPL was statistically significant, with F = 17.26, df = 1, 23, P < 0.0004,  $r^2 = 0.43$ , and residual mean square error = 0.21.

Male song bioassays. The mean percentage captured in the trap next to broadcast calling song  $(67 \pm 2\%)$  was significantly higher than the mean in the silent control  $(43 \pm 3)$ , with F = 16.7, df = 3, 86, P < 0.001, and minimum significant difference (MSD) = 8.1 using the Waller-Duncan k-ratio t-test.

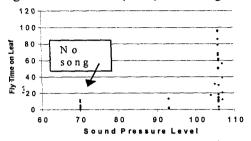


FIGURE 1. Mean fly time on leaf for virgin female medflies exposed to male song at different sound levels

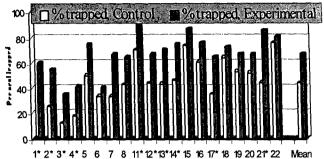


FIGURE 2. Percentages of virgin female medflies captured in yellow adhesive traps positioned near experimental (sound) or control (no sound) speaker/funnel devices at opposite ends of bioassay chamber.

In summary, broadcast male calling song was found to significantly affect female medfly behavior. However, the tested song did not induce phonotaxis, and it affected female behavior over a relatively short, <0.5-m distance, even at high signal levels. Further increases in attractivity also may result from addition of olfactory (pheromonal stimuli) or visual attractants that mimic the environments where leks form most frequently (10) are also being considered as ways to increase the range of the trap while maintaining selectivity.

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